abcam

ab251187

Anti-EHMT2/G9A+EHMT1/GLP antibody [EPR18667] - BSA and Azide free

Recombinant | RabMAb

Key facts

Isotype	IgG
Host species	Rabbit
Storage buffer	pH: 7.2 - 7.4 Constituents: PBS
Form	Liquid
Clonality	Monoclonal
Immunogen	The exact immunogen used to generate this antibody is proprietary information.
Carrier free	Yes
Clone number	EPR18667

Reactivity data

ICC/IF

Tested

Species	Mouse, Human
Dilution info	-
Notes	-

Expected

Species	Rat
Dilution info	Use at an assay dependent concentration.
Notes	-

WB

Tested

Species	Rat, Mouse, Human
Dilution info	-
Notes	-

Flow Cyt (Intra)

Tested

Species	Human
Dilution info	-
Notes	-

Expected

Species	Mouse, Rat
Dilution info	Use at an assay dependent concentration.
Notes	-

IHC-P

Tested

Species	Human
Dilution info	-
Notes	Perform heat-mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

Expected

Species	Mouse, Rat
Dilution info	Use at an assay dependent concentration.
Notes	-

IP

Not recommended

Species	Human, Mouse, Rat
Dilution info	-
Notes	-

Target data

See full target information EHMT1

Function

Histone methyltransferase that specifically mono-, di- and trimethylates 'Lys-9' of histone H3 (H3K9me1, H3K9me2 and H3K9me3, respectively) in euchromatin (PubMed:12004135). H3K9me represents a specific tag for epigenetic transcriptional repression by recruiting HP1 proteins to methylated histones (PubMed:12004135). Also weakly methylates 'Lys-27' of histone H3 (H3K27me) (PubMed:12004135). Also required for DNA methylation, the histone methyltransferase activity is not required for DNA methylation, suggesting that these 2 activities function independently (By similarity). Probably targeted to histone H3 by different DNA-binding proteins like E2F6, MGA, MAX and/or DP1 (PubMed:12004135). During G0 phase, it probably contributes to silencing of MYC- and E2F-responsive genes, suggesting a role in G0/G1 transition in cell cycle (PubMed:12004135). Involved in the differentiation of myoblastic precursors into brown adipose cells: following recruitment to chromatin by PRDM16, mediates formation of H3K9me2 and H3K9me3, inhibiting the expression of white adipose-selective genes (By similarity). Also involved in the differentiation of beige adipocytes from white adipose cells following recruitment by PRDM16 (By similarity). EHMT1 also promotes protein stabilization of PRDM16, by preventing PRDM16 ubiquitination and degradation (By similarity). In addition to the histone methyltransferase activity, also methylates non-histone proteins: mediates dimethylation of 'Lys-373' of p53/TP53 (PubMed:20118233). Represses the expression of mitochondrial function-related genes, perhaps by occupying their promoter regions, working in concert with probable chromatin reader BAZ2B (By similarity).

Storage

Shipped at conditions	Blue Ice

Appropriate short-term storage conditions	+4°C
Appropriate long-term storage conditions	+4°C
Storage information	Do Not Freeze

Notes

ab251187 is the carrier-free version of ab194299.

Patented technology
Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMAb® patents.

What are the advantages of a recombinant monoclonal antibody?
This product is a recombinant monoclonal antibody, which offers several advantages including:

- High batch-to-batch consistency and reproducibility
- Improved sensitivity and specificity
- Long-term security of supply
- Animal-free batch production

For more information, read more on recombinant antibodies.

Conjugation ready
Our carrier-free antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. This conjugation-ready format is designed for use with fluorochromes, metal isotopes, oligonucleotides, and enzymes, which makes them ideal for antibody labelling, functional and cell-based assays, flow-based assays (e.g. mass cytometry) and Multiplex Imaging applications.

Use our conjugation kits for antibody conjugates that are ready-to-use in as little as 20 minutes with 1 minute hands-on-time and 100% antibody recovery: available for fluorescent dyes, HRP, biotin and gold.

Compatibility
This product is compatible with the Maxpar® Antibody Labeling Kit from Fluidigm, without the need for antibody preparation. Maxpar® is a trademark of Fluidigm Canada Inc.

Supplementary info

This supplementary information is collated from multiple sources and compiled automatically.

Activity summary	EHMT2 also known as G9a and EHMT1 referred to as GLP are lysine methyltransferases that add methyl groups to histone H3 on lysine 9 (H3K9). The functional proteins exist as dimers comprising a catalytic SET domain that mediates their enzymatic activity. EHMT2 has a mass of approximately 150 kDa while EHMT1 is about 143 kDa. These proteins are expressed in various tissues but show high expression levels in the brain liver and placenta. They play an essential role in chromatin modification which impacts gene expression.

Biological function summary	EHMT2 and EHMT1 form an important protein complex responsible for histone methylation. This complex regulates gene silencing through the tight modulation of chromatin structure affecting cellular processes such as differentiation and proliferation. EHMT2/G9a and EHMT1/GLP participate in establishing and maintaining transcriptional repression during development. The complex works in concert with other chromatin modifiers to control gene expression patterns contributing to the cellular and tissue-specific transcriptional programs.
Pathways	The functions of EHMT2 and EHMT1 integrate into the gene silencing pathways including the PRC2-mediated histone modification pathway. Within these pathways proteins like EZH2 a member of the Polycomb repressive complex 2 regulate histone methylation state further influencing gene expression patterns. EHMT2/G9a and EHMT1/GLP link signal transduction pathways to chromatin remodeling enabling cells to adapt their transcriptional activity according to external signals and developmental cues.
Associated diseases and disorders	Mutations or dysregulation of EHMT2 and EHMT1 have associations with neurological disorders like Kleefstra syndrome and certain cancers such as acute myeloid leukemia. The proteins' involvement in these disorders connects with other proteins like G9a and GLP through epigenetic silencing mechanisms emphasizing their influence on development and disease progression. Targeting EHMT2/G9a and EHMT1/GLP pathways can have potential therapeutic implications for treating these conditions highlighting the importance of understanding their functional roles in health and disease.

Product promise

Tested

We have tested this species and application combination and it works. It is covered by our product promise.

Expected

We have not tested this specific species and application combination in-house, but expect it will work. It is covered by our product promise.

Predicted

This species and application combination has not been tested, but we predict it will work based on strong homology. However, this combination is not covered by our product promise.

Not recommended

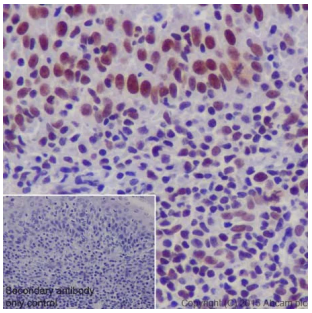
We do not recommend this combination. It is not covered by our product promise.

We are dedicated to supporting your work with high quality reagents and we are here for you every step of the way should you need us.

In the unlikely event of one of our products not working as expected, you are covered by our product promise.

Full details and terms and conditions can be found here:

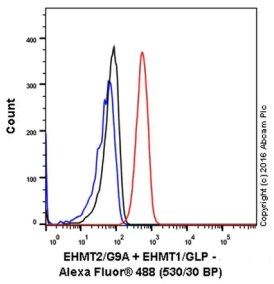
10 product images



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-EHMT2/G9A+EHMT1/GLP antibody [EPR18667] - BSA and Azide free (ab251187)

EHMT2/G9A+EHMT1/GLP Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-EHMT2/G9A+EHMT1/GLP antibody

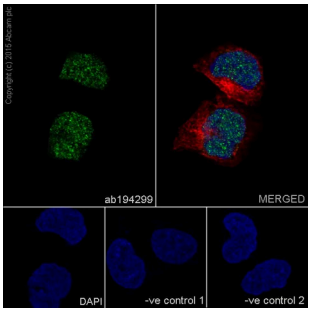
This data was developed using ab194299, the same antibody clone in a different buffer formulation. Immunohistochemical analysis of paraffin-embedded human tonsil tissue labeling EHMT2/G9A + EHMT1/GLP with ab194299 at 1/1000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/500 dilution. Nuclear staining on human tonsil is observed. Counter stained with Hematoxylin. Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/500 dilution. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Flow Cytometry (Intracellular) - Anti-EHMT2/G9A+EHMT1/GLP antibody [EPR18667] - BSA and Azide free (ab251187)

EHMT2/G9A+EHMT1/GLP Flow Cytometry (Intracellular) staining using rabbit Anti-EHMT2/G9A+EHMT1/GLP antibody

This data was developed using ab194299, the same antibody clone in a different buffer formulation.
Intracellular Flow Cytometry analysis of HeLa (human cervix adenocarcinoma) cells labelling EHMT2/G9A + EHMT1/GLP (red) with purified ab194299 at a dilution of 1/150. Goat anti rabbit IgG (Alexa Fluor® 488) was used as the secondary antibody at 1/2000. Cells were fixed with 4% paraformaldehyde and permeabilised with 90% methanol. Isotype control antibody was Rabbit monoclonal IgG (black). The blue line shows cells without incubation with primary antibody and secondary antibody.

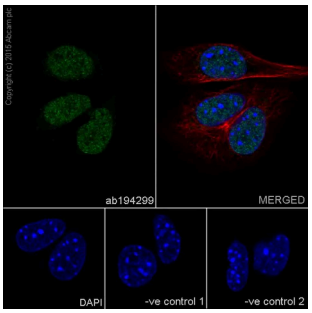


Immunocytochemistry/ Immunofluorescence - Anti-EHMT2/G9A+EHMT1/GLP antibody [EPR18667] - BSA and Azide free (ab251187)

EHMT2/G9A+EHMT1/GLP Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-EHMT2/G9A+EHMT1/GLP antibody

This data was developed using ab194299, the same antibody clone in a different buffer formulation. Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized HeLa (Human epithelial cell line from cervix adenocarcinoma) cells labeling EHMT2/G9A + EHMT1/GLP with ab194299 at 1/1000 dilution, followed by Goat Anti-Rabbit IgG (Alexa Fluor® 488) (ab150077) secondary antibody at 1/1000 dilution (green). Confocal image showing nuclear staining on HeLa cell line. The nuclear counter stain is DAPI (blue). Tubulin is detected with Anti-alpha Tubulin mouse MAb (ab7291) at 1/1000 dilution, followed by Goat Anti-Mouse IgG H&L (Alexa Fluor® 594) (ab150120) secondary antibody at 1/1000 dilution (red). The negative controls are as follows:

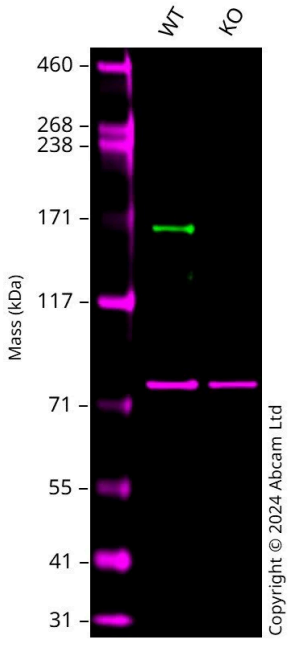
-ve control 1: ab194299 at 1/1000 dilution, followed by Goat Anti-Mouse IgG H&L (Alexa Fluor® 594) (ab150120) at 1/1000 dilution.
-ve control 2: Anti-alpha Tubulin mouse MAb (ab7291) at 1/1000 dilution, followed by Goat Anti-Rabbit IgG (Alexa Fluor® 488) (ab150077) at 1/1000 dilution.



Immunocytochemistry/ Immunofluorescence - Anti-EHMT2/G9A+EHMT1/GLP antibody [EPR18667] - BSA and Azide free (ab251187)

EHMT2/G9A+EHMT1/GLP Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-EHMT2/G9A+EHMT1/GLP antibody

This data was developed using ab194299, the same antibody clone in a different buffer formulation.Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% tritonX-100 permeabilized NIH/3T3 (Mouse embryonic fibroblast cell line) cells labeling EHMT1/GLP + EHMT2/G9A with ab194299 at 1/1000 dilution, followed by Goat Anti-Rabbit IgG (Alexa Fluor® 488) (ab150077) secondary antibody at 1/1000 dilution (green). Confocal image showing nuclear staining on NIH/3T3. The nuclear counter stain is DAPI (blue).Tubulin is detected with Anti-alpha Tubulin mouse MAb (ab7291) at 1/1000 dilution, followed by Goat Anti-Mouse IgG H&L (Alexa Fluor® 594) (ab150120) secondary antibody at 1/1000 dilution (red).
The negative controls are as follows:
-ve control 1: ab194299 at 1/1000 dilution, followed by Goat Anti-Mouse IgG H&L (Alexa Fluor® 594) (ab150120) secondary at 1/1000 dilution.
-ve control 2: Anti-alpha Tubulin mouse MAb (ab7291) at 1/1000 dilution, followed by Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (ab150077) secondary at 1/1000 dilution.



Western blot - Anti-EHMT2/G9A+EHMT1/GLP antibody [EPR18667] - BSA and Azide free (ab251187)

This data was developed using the same antibody clone in a different buffer formulation (ab194299).

Western blot: Anti-EHMT1 antibody [EPR18667] (ab194299) staining at 1/1000 dilution, shown in green; Mouse anti-CANX [CANX/1543] (ab238078) loading control staining at 1/20000 dilution, shown in magenta. In Western blot, ab194299 was shown to bind specifically to EHMT1. A band was observed at 165 kDa in wild-type HCT 116 cell lysates with no signal observed at this size in EHMT1 knockout cell line. To generate this image, wild-type and EHMT1 knockout HCT 116 cell lysates were analysed. First, samples were run on an SDS-PAGE gel then transferred onto a nitrocellulose membrane. Membranes were blocked in 3 % milk in TBS-0.1 % Tween® 20 (TBS-T) before incubation with primary antibodies overnight at 4 °C. Blots were washed four times in TBS-T, incubated with secondary antibodies for 1 h at room temperature, washed again four times then imaged. Secondary antibodies used were Goat anti-Rabbit IgG H&L 800CW and Goat anti-Mouse IgG H&L 680RD at 1/20000 dilution.
All lanes:
Western blot - Anti-EHMT2/G9A + EHMT1/GLP antibody [EPR18667] (ab194299) at 1/1000 dilution

Lane 1:
Wild-type HCT 116 cell lysate at 20 µg

Lane 2:
EHMT1 knockout HCT 116 cell lysate at 20 µg

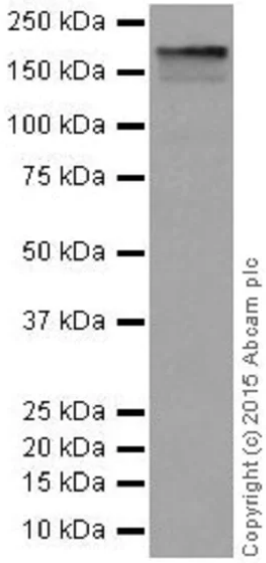
Lane 2:
Western blot - Human EHMT1 knockout HCT116 cell line (ab287388)

Secondary

All lanes:
Goat anti-Rabbit IgG H&L 800CW and Goat anti-Mouse IgG H&L 680RD at 1/20000 dilution

Performed under reducing conditions.

Observed band size: 165 kDa



Western blot - Anti-EHMT2/G9A+EHMT1/GLP antibody [EPR18667] - BSA and Azide free (ab251187)

EHMT2/G9A+EHMT1/GLP Western blot staining of Human fetal kidney lysate using rabbit Anti-EHMT2/G9A+EHMT1/GLP antibody

This data was developed using ab194299, the same antibody clone in a different buffer formulation.
Blocking and dilution buffer: 5% NFDM/TBST.

All lanes:
Western blot - Anti-EHMT2/G9A + EHMT1/GLP antibody [EPR18667] (ab194299) at 1/1000 dilution

All lanes:
Human fetal kidney lysate at 10 µg

Secondary

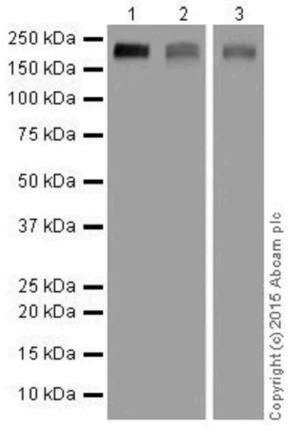
All lanes:
Anti-Rabbit IgG (HRP), specific to the non-reduced form of IgG at 1/10000 dilution

Developed using the ECL technique.

Predicted band size: 132 kDa, 141 kDa

Observed band size: 170 kDa

Exposure time: 3min



Western blot - Anti-EHMT2/G9A+EHMT1/GLP antibody [EPR18667] - BSA and Azide free (ab251187)

EHMT2/G9A+EHMT1/GLP Western blot staining using rabbit Anti-EHMT2/G9A+EHMT1/GLP antibody

This data was developed using ab194299, the same antibody clone in a different buffer formulation.
Blocking and dilution buffer: 5% NFDM/TBST.

Exposure times: Lanes 1-2: 20 seconds; Lane 3: 30 seconds.

All lanes:
Western blot - Anti-EHMT2/G9A + EHMT1/GLP antibody [EPR18667] (ab194299) at 1/5000 dilution

Lane 1:
HEK-293 (Human epithelial cell line from embryonic kidney) whole cell lysate at 10 µg

Lane 2:
HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate at 10 µg

Lane 3:

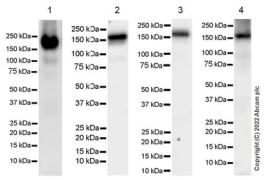
K562 (Human chronic myelogenous leukemia cell line from bone marrow) whole cell lysate at 10 µg

Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/100000 dilution

Predicted band size: 132 kDa, 136 kDa, 141 kDa

Observed band size: 136 kDa, 170 kDa



Western blot - Anti-EHMT2/G9A+EHMT1/GLP antibody [EPR18667] - BSA and Azide free (ab251187)

Blocking buffer and concentration: 5% NFDM/TBST Diluting buffer and concentration: 5% NFDM/TBST Exposure Time: Lane 1: 15 seconds, Lane 2-4: 37 seconds. Lysates were freshly made and used for Western blotting immediately to minimize protein degradation. This data was developed using ab194299, the same antibody clone in a different buffer formulation.
All lanes:
Western blot - Anti-EHMT2/G9A + EHMT1/GLP antibody [EPR18667] (ab194299) at 1/1000 dilution

Lane 1:
HeLa (human cervix adenocarcinoma epithelial cell) whole cell lysate at 20 µg

Lane 2:
NIH/3T3 (mouse embryonic fibroblast) whole cell lysate at 20 µg

Lane 3:
PC-12 (rat adrenal gland pheochromocytoma cell) whole cell lysate at 20 µg

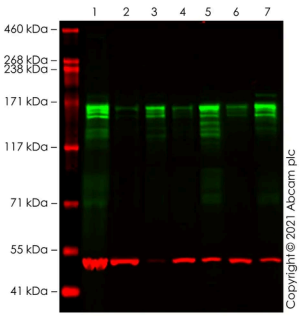
Lane 4:
Raw 264.7 (mouse Abelson murine leukemia virus-induced tumor macrophage) whole cell lysate at 20 µg

Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/100000 dilution

Predicted band size: 141 kDa, 132 kDa

Observed band size: 170 kDa



Western blot - Anti-EHMT2/G9A+EHMT1/GLP antibody [EPR18667] - BSA and Azide free (ab251187)

EHMT2/G9A+EHMT1/GLP Western blot staining using rabbit Anti-EHMT2/G9A+EHMT1/GLP antibody

False colour image of Western blot: Anti-EHMT2/G9A + EHMT1/GLP antibody [EPR18667] staining at 1/1000 dilution, shown in green; Mouse anti-Alpha Tubulin [DM1A] (ab7291) loading control staining at 1/20000 dilution, shown in red. In Western blot, ab194299 was shown to bind specifically to EHMT1/GLP and EHMT2/G9A. First, samples were run on an SDS-PAGE gel then transferred onto a nitrocellulose membrane. Membranes were blocked in 3 % milk in TBS-0.1 % Tween® 20 (TBS-T) before incubation with primary antibodies overnight at 4°C. Blots were washed four times in TBS-T, incubated with secondary antibodies for 1 h at room temperature, washed

again four times, then imaged. Secondary antibodies used were Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed (ab216773) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed (ab216776) at 1/20000 dilution.
All lanes:
Western blot - Anti-EHMT2/G9A + EHMT1/GLP antibody [EPR18667] (ab194299) at 1/1000 dilution

Lane 1:
Wild-type HAP1 cell lysate at 40 µg

Lane 2:
EHMT1 knockout HAP1 cell lysate at 40 µg

Lane 3:
Wild-type HAP1 Nuclear cell lysate at 40 µg

Lane 4:
HeLa cell lysate at 40 µg

Lane 5:
HeLa Nuclear cell lysate at 40 µg

Lane 6:
Wild-type HEK-293T cell lysate at 40 µg

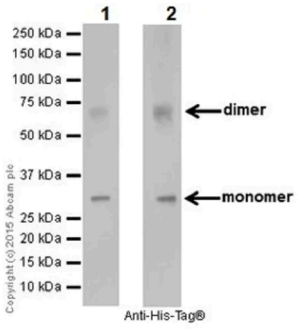
Lane 7:
Wild-type HEK-293T nuclear cell lysate at 40 µg

Secondary

All lanes:
Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed (ab216773) and Goat anti-Mouse IgG H&L (IRDye®680RD) preabsorbed (ab216776) at 1/20000 dilution

Predicted band size: 132 kDa, 141 kDa

Observed band size: 150-170 kDa



Western blot - Anti-EHMT2/G9A+EHMT1/GLP antibody [EPR18667] - BSA and Azide free (ab251187)

This data was developed using ab194299, the same antibody clone in a different buffer formulation.
Blocking and dilution buffer: 5% NFDM/TBST.

EHMT2 recombinant protein fragment with His-Tag®.

Lane 1:
Western blot - Anti-EHMT2/G9A + EHMT1/GLP antibody [EPR18667] (ab194299) at 1/5000 dilution

Lane 2:
Anti His-Tag®.

All lanes:
EHMT2 recombinant protein at 0.01 µg

Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/100000 dilution

Developed using the ECL technique.

Predicted band size: 132 kDa, 141 kDa

Observed band size: 30 kDa, 60 kDa

Exposure time: 5s

Please note: All products are 'FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC OR THERAPEUTIC PROCEDURES'.